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ANATOMY OF THE REPRODUCTIVE SYSTEM IN SIX *ANASTREPHA* SPECIES AND COMMENTS REGARDING THEIR TERMINOLOGY IN TEPHRITIDAE (DIPTERA)

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Abstract.—A comparative study of the anatomy of the reproductive system of mature males and females of the following species was performed: *A. serpentina* (Wiedemann), *A striata* Schiner, *A cordata* Aldrich, *A. ludens* (Loew), *A. obliqua* (Macquart) and *A. fraterculus* (Wiedemann). The study material came from diverse regions of the states of Veracruz and Chiapas, Mexico. In females, several important differences among species were observed: in the number of ovarioles, morphology of the ventral receptacle, *signum,* spermathecae and aculeus. In males, differences were apparent in the accessory glands, ejaculatory apodeme of the sperm pump, distiphallus and surstyli. For the first time, the real location of the seminal vesicles is shown and the aedeagal gland is described. A comparative discussion ensues regarding terminology and the location of various structures in both sexes.

Key Words: *Anastrepha,* reproductive system, females, males, anatomy

The genus *Anastrepha* includes 185 species occurring only in the Western Hemisphere. Despite the great diversity and wide distribution of this genus, most studies have concentrated exclusively on a few species of economic importance (Hernández-Ortiz and Aluja 1993).

Williamson's (1989) review and synthesis of studies of the descriptive anatomy of the reproductive system, oogenesis and spermatogenesis was limited to only a few fruit fly species. Studies of females of the following species have been published: *Rhagoletis pomonella* (Walsh) (Dean 1935), *Ceratitis capitata* (Wiedemann) (Hanna 1938, Valdéz Carrasco and Prado Beltrán 1990), *Xanthaciura unipuncta* Malloch (Souza Lopes 1939), *Bactrocera tryoni* (Froggatt) (formerly *Strumeta*) (Drew 1969), *Bactrocera oleae* Gmelin (formerly *Dacus*) (Solinas and Nuzzaci 1984), as well as *Rhagoletis nova* (Schiner) and *R. conversa* (Brethes) (Flores et al. 1987).

For *Anastrepha* there is only general information regarding the anatomy of the reproductive system in females for *A. ludens* (Loew) (Dampf 1933, Servín-Villegas and Jiménez-Jiménez 1995), for *A. suspensa* (Loew) (Dodson 1978) and for *A. serpentina* (Wiedemann) (Martínez et al. 1995).

With respect to males, the information presently available consists of studies in *C. capitata* (Hanna 1938, Anwar et al., 1971), *X. unipuncta* (Souza Lopes 1939) and *B. tryoni* (Drew 1969). The studies of *Anastrepha* include species such as the following: *A. suspensa* (Dodson 1978), *A. serpentina* (Martínez et al. 1995), *A. ludens* (Ser-

vín-Villegas and Jiménez-Jiménez 1995) and several unidentified *Anastrepha* species (Bressan 1995).

A review of the aforementioned literature shows that much of the terminology used for Tephritidae by these authors differs substantially, regarding names and locations of certain structures.

In this paper we present a comparative study of the anatomy of the reproductive system for males and females of the following species: *A. serpentina* (Wiedemann), *A. striata* Schiner, *A. cordata* Aldrich, *A. ludens* (Loew), *A. obliqua* (Macquart) and *A. fraterculus* (Wiedemann). Furthermore, terminology and the location of certain structures such as the seminal vesicles and the aedeagal gland in males, and the *bursa copulatrix* and the ventral receptacle in females are discussed.

Material and Methods

Males and females of six *Anastrepha* species were obtained from larvae infesting their natural host plants from the following regions: *A. serpentina* from Región del Soconusco, Chiapas reared in *Pouteria sapota* (Sapotaceae); *A. striata* and *A. fraterculus* from the "Estación de Biología Tropical Los Tuxtlas", Veracruz reared in *Psidium guajava* (Myrtaceae); *A. cordata* from the "Estación de Biología Tropical Los Tuxtlas", reared in *Tabernaemontana alba* (Apocynaceae); *A. ludens* from Martínez de la Torre, Veracruz, reared in *Citrus sinensis* (Rutaceae); and *A. obliqua* from Apazapan, Veracruz, reared in *Spondias purpurea* (Anacardiaceae).

The emergence and development of adults took place under laboratory conditions; until the gonads reached full maturity, hydrolized protein (ICN-Biomedial Inc.) was provided as a food source. Approximately 50 individuals of each species and sex were analyzed (except *A. cordata,* for which only 7 specimens were observed).

Dissection of the individuals was performed under a stereomicroscope. Each specimen was placed in a petri dish in Ringer's solution. The reproductive systems were drawn while in this solution using a camera lucida. Specimens were fixed on slides in Carnoy's solution, then dehydrated, cleared and stained using the Feulgen light green technique. Finally, they were mounted whole in Canada Balsam (Gabe 1968).

In order to observe particular morphological structures such as the cuticular intima of the vaginal duct, ventral receptacle, spermathecae, and the aculeus in females, as well as the vas deferens, accessory glands and sperm pump in males, some reproductive systems were dissected in NaOH solution (4%) and stained with chlorazol black E following the technique described by Carayon (1969). Later they were dehydrated, cleared and mounted in Canada Balsam.

The terminalia of both sexes were prepared by cutting open the entire abdomen in NaOH (10%) solution which was boiled for a few minutes in order to remove all extraneous materials; the terminalia were later analyzed with a compound microscope. Microphotographs were made with a Zeiss photomicroscope.

Results

Female Reproductive System

The general terminology employed here to describe females is based on the works of McAlpine (1981) and Norrbom and Kim (1988), and for particular internal structures we following Dean (1935). There is considerable terminological confusion among later studies that used different anatomical terms for the same structure or used incorrect terminology (Table 1).

The female *Anastrepha* reproductive system is made up of the following structures: two ovaries, two lateral oviducts, the common oviduct, two accessory glands, three spermathecae with their respective ducts, vagina and aculeus (Fig. 1A).

Even in mature females, the ovary is covered by a thin wall. The ovarioles (Fig.

Table 1. Comparison of some terminology used for the female reproductive system in Tephritidae.

Present Study	Dampf (1933)	Dean (1935)	Hanna (1938)	Drew (1969)	Solinas & Nuzzaci (1984)	Williamson (1989)
Ovary	ovary	ovary	ovary	ovary	ovary	ovary
Lateral oviduct	not indicated	lateral oviduct	oviduct	lateral oviduct	lateral oviduct	lateral oviduct
Common oviduct	not indicated	median oviduct	common oviduct	not indicated	common oviduct	common oviduct
Accessory glands	accessory glands	accessory glands	colleterial glands	colleterial glands	accessory glands	accessory glands
Spermathecae	spermathecae	spermathecae	spermathecae	spermathecae	receptaculum seminis	spermathecae
Bursa copulatrix	not indicated	*bursa copulatrix*	vagina	vagina	anterior vagina	*bursa copulatrix*
Ventral receptacle	not indicated	ventral receptacle	vagina	morula gland	fertilization chamber	ventral receptacle
Vaginal duct	oviduct	vagina	vaginal duct	vaginal duct	posterior vagina	vaginal duct
Cloacal opening	end oviduct	cloaca	genital opening	gonopore	not indicated	vulva

1B) are of the polytrophic type and have a terminal filament, germarium, vitellarium and calyx. The germarium contains the oögonial cells, nutritive cells and prefollicular cells. The follicles found in the vitellarium show varying degrees of maturity; each one is formed by 16 cells of germinal origin corresponding to one oocyte and 15 nutritive cells which are contained in the cavity defined by the follicular epithelium. The calyx, which opens into its respective lateral oviduct, is a prolongation of the vitellarium. The lateral oviducts are short and their walls very thin, both open into the common oviduct, which is slightly longer and reaches the anterior part of the vagina.

The vagina has two regions with different characteristics. The anterior part is modified to form a differentiated region of distinctive structure called the *bursa copulatrix.* The posterior part or vaginal duct is a long tubular duct which folds up before penetrating the aculeus. Parallel to the rectum, it flows into the cloaca.

The *bursa copulatrix* can be observed externally due to the greater thickness of its wall and because both the spermathecal and accessory gland ducts open into its dorsal side. The thickness of the *bursa copulatrix* is partly due to the presence of the ventral receptacle, a flatttened, sac-like structure protruding cephalad from its ventral side. The ventral receptacle has thick, highly ornamented cuticular walls.

The vaginal duct is long and tubular and it has a number of longitudinal pleats. At the end of its basal third, positioned dorsally, are two cuticular pieces called the *signum.*

Both accessory glands are oval in shape, voluminous and opaque. Each gland has a bulky base and extends into a thin, long duct which opens into the anterior part of the vagina.

The spermathecae are each made up of a sclerotized capsule surrounded by a secretory epithelium connected by a very long and thin spermathecal duct. All ducts open into the dorsal side of the *bursa copulatrix.*

The aculeus is a highly sclerotized structure; its morphology and length differ among species of *Anastrepha,* making it extremely important for the taxonomy of the genus.

In the studied *Anastrepha* species, the female reproductive system varies mainly in the number of ovarioles and in the morphological characteristics of the ventral receptacle, as well as in the *signum,* spermathecae and aculeus.

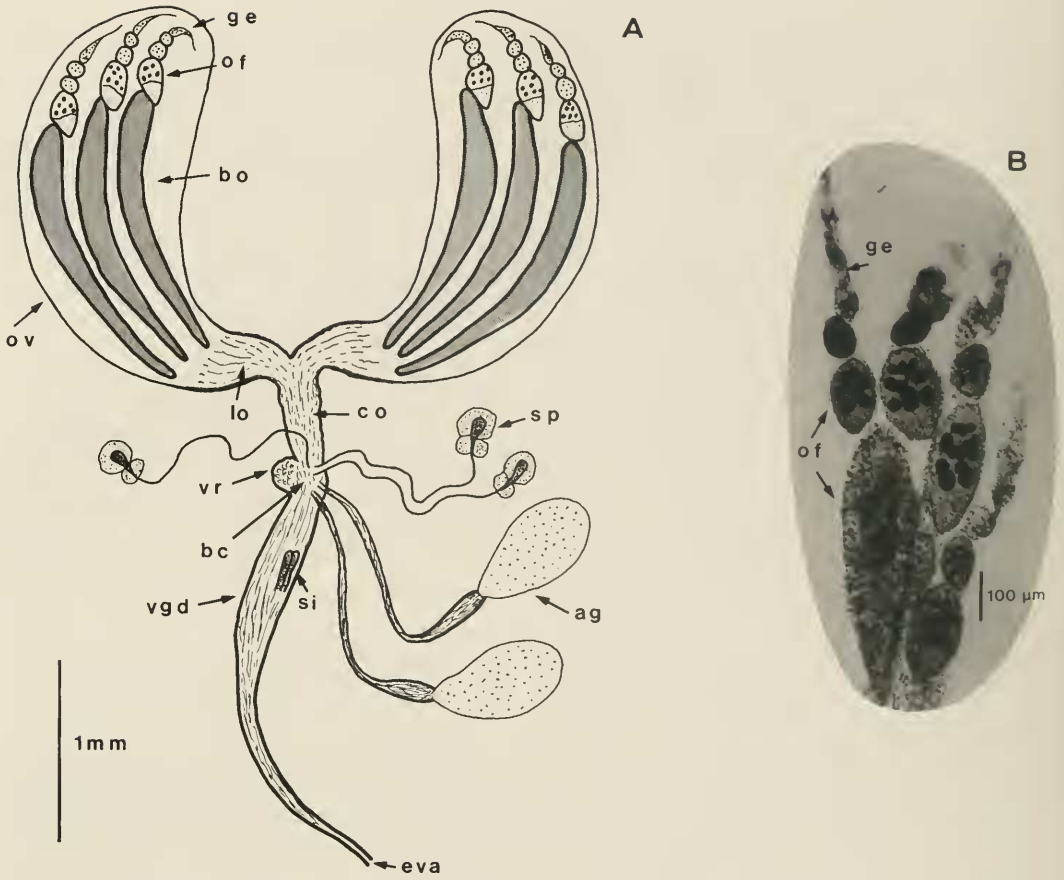

Fig. 1. Reproductive system of the female of *Anastrepha serpentina*. A, General morphology of the reproductive system. B, Microphotograph of the apical end of some ovarioles. Abbreviations: ag = accessory gland; bc = *bursa copulatrix*; bo = basal mature oocyte; co = common oviduct; eva = entrance of the vagina to aculeus; ge = germarium; lo = lateral oviduct; of = ovarian follicles in the vitellarium; ov = ovary (with some ovarioles figured); si = signum; sp = spermatheca; vgd = vaginal duct; vr = ventral receptacle.

The number of ovarioles present in the six species varied between 20 and 33 ovarioles per ovary. The highest average was observed in *A. obliqua* (30–33), *A. serpentina* (25–30) and *A. cordata* (25–28); the lowest averages were in *A. fraterculus* (20–24), *A. striata* (20–25) and *A. ludens* (22–25) (Table 2).

The ventral receptacle is usually elongated with numerous lobular papillae on its surface (Fig. 2A,B,D,E,F) except in *A. cordata,* in which it is almost spherical, shorter and almost completely without papillae (Fig. 2C; Table 2).

The *signum* is formed by two sclerotized plates which are long and sclerotized in *A. serpentina* and *A. striata* (Fig. 2A', B'), short and less sclerotized in *A. ludens* (Fig. 2D',E'), very short and more or less sclerotized in *A. fraterculus* (Fig. 2F') and inconspicuous in *A. cordata* (Fig. 2C').

The glandular units of the spermathecal secretory epithelium differ among species (Fig. 3). The spermathecal capsules are spherical in shape and usually covered by numerous sclerotized spiculae. The spermathecae of *A. ludens* tend to be longer than those of other species. In *A. cordata,* in contrast, the spermathecae are rounded and the spiculae are lacking. In *A. frater-*

Table 2. Some comparative characteristics in females of the *Anastrepha* species examined during this study.

Species	Number Ovarioles	Ventral Receptacle	*signum*
A. serpentina	25–30	elongated	2 plates
A. striata	20–25	elongated	2 plates
A. cordata	25–28	short	inconspicuous
A. ludens	22–25	elongated	2 plates
A. obliqua	30–33	elongated	2 plates
A. fraterculus	20–24	elongated	2 plates

culus the spermathecae are not consistently rounded, as they are in *A.obliqua* (Fig. 4 A–F).

The aculeus is usually sclerotized and dorsoventrally compressed. In *A. serpentina* it measures from 2.9–3.3 mm long; the tip (Fig. 4A') has tiny teeth which occupy the apical third. In *A. striata,* the aculeus is notably robust, measuring 2.1–2.3 mm long; its tip (Fig.4B') is broad with a marked constriction after the cloacal opening. The aculeus of *A. cordata* is very thin (less than 0.09 mm wide) and measures 4.7–5.5 mm long; the tip is short and lacks teeth laterally (Fig. 4C').

In species belonging to the *fraterculus* group, differences were observed in the length of aculeus tip and shape of its teeth. The aculeus of *A. ludens* varies in length from 3.2–5.0 mm and has scant broad, rounded teeth which occupy less than the apical half. In *A. fraterculus* it measures from 1.6–1.8 mm and has large, rounded teeth occupying half of the apical end; while in *A. obliqua* it measures from 1.4–1.7 mm and has large, sharp teeth which occupy three-fourths of the apical end (Figs. 4 D'–E'–F').

Male Reproductive System

The general terminology used to describe the male reproductive system is based on the works of Matzuda (1976) and McAlpine (1981), whereas for particular internal structures we followed Hanna (1938). We have also included a comparative synopsis of the terms employed for *Anastrepha* by some other authors (Table 3).

The male reproductive system is made up of two testes, two vas deferens, an ejaculatory duct, two seminal vesicles, several pairs of accessory glands, a sperm pump, an aedeagal gland and the aedeagus itself (Fig. 5).

The testes are oval-shaped and intensely bright yellow. In the apical part of each testis there is a germarium followed by regions of primary spermatocytes, secondary spermatocytes, spermatids, and sperm bundles, beyond which lies the basal region which, in mature males, contains a large quantity of free spermatozoids (Fig. 6A). Each testis opens into a vas deferens characterized by a very broad anterior part which thins out further on, finally opening into the anterior part of the ejaculatory duct.

In the ejaculatory duct, which is very long, three regions can be distinguished: the widest, the anterior region, into which two vas deferens, two seminal vesicles, and various pairs of accessory glands open; the intermediate region, which has a smaller diameter than the first and which, extremely thin at the end, runs through the base of the sperm pump; and the posterior region, which begins beyond the sperm pump and has a smaller diameter than the former two regions. Most of the last region is covered by a thick pleated wall, which makes its diameter look bigger; however, the external wall disappears distally immediately before reaching the basiphallus. At precisely this point, it connects to the aedeagal gland.

The accessory glands are paired, long and translucid; their wall is slightly thicker than the seminal vesicles.

There are two elongated seminal vesicles with thin walls; free spermatozoids, apparently immersed in a secretion, are visible in their interior. The vesicles open into the anterior part of the ejaculatory duct parallel to the vas deferens (Fig. 6 B–C).

The sperm pump is pear-shaped with a narrow base; it is composed of two highly sclerotized structures of a cuticular nature.

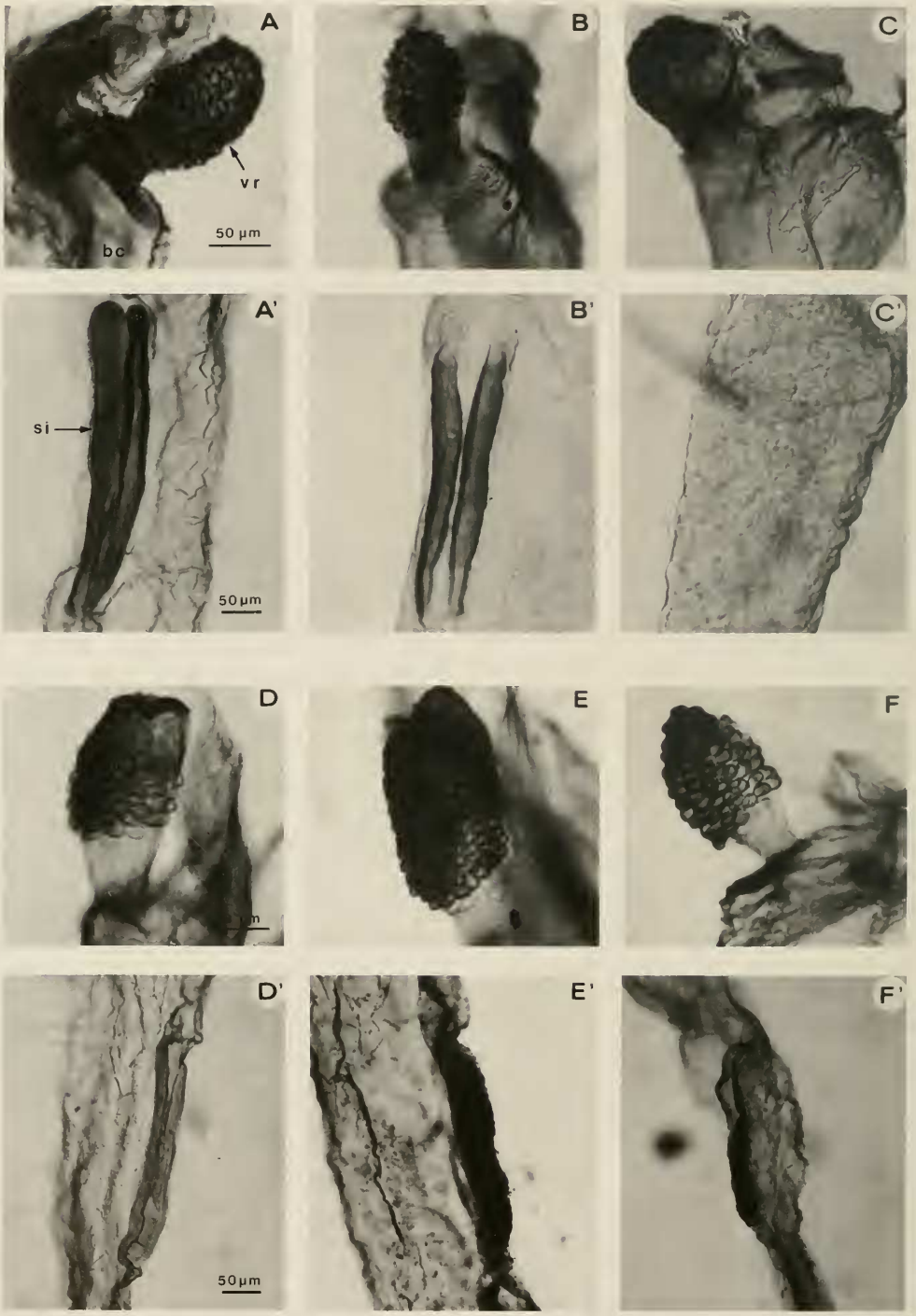

Fig. 2. Microphotographs of the cuticular intima of the *bursa copulatrix* showing the ventral receptacle and area of the vaginal duct near the signum (clorazol black E). A, A', *Anastrepha serpentina*. B, B', *A. striata*. C, C', *A. cordata*. D, D', *A. ludens*. E, E', *A. obliqua*. F, F', *A. fraterculus*. Abbreviations: bc = bursa copulatrix; co = common oviduct; si = signum; vr = ventral receptacle.

Table 3. Comparison of some terminology used for the male reproductive system in the Tephritidae.

Present Study	Hanna (1938)	Drew (1969)	Dodson (1978)	Williamson (1989)	Bressan (1995)	Martínez et al (1995)
Testis	testis	testis	testis	testis	testis	testis
Vas deferens	vas deferens	vas deferens	vas deferens	vas deferens	vas deferens	vas deferens
Ejaculatory duct	vas efferents	ejaculatory duct	common duct	ejaculatory duct	vas efferents	ejaculatory duct
Accessory glands	accessory glands	accessory glands	accessory glands	accessory glands	accessory glands	accessory glands
Seminal vesicles	end of vas deferens	not indicated	not indicated	base of testis	base of testis	not indicated
Sperm pump	pump organ	pump organ	sperm pump	ejaculatory apodeme	ejaculatory apodeme	ejaculatory apodeme
Aedeagal gland	gland	oval gland	not indicated	not indicated	2nd accessory gland	not indicated
Aedeagus	aedeagus	aedeagus	aedeagus	aedeagus	aedeagus	not indicated
Distiphallus	not indicated	not indicated	terminal end	not indicated	not indicated	not indicated

The ejaculatory apodeme is long and spatula-shaped with a thick layer of muscles inserted onto it longitudinally. The other structure is capsule-like and semi-spherical; it surrounds the ejaculatory duct and the ejaculatory apodeme inserts into it (Fig. 6 D). Due to the complexity of this organ, it is very difficult to determine the exact position of the ejaculatory duct.

The aedeagal gland is an unpaired, elongated, and usually voluminous organ. It is formed by glandular units which surround the cuticular glandular reservoir. The basal region of the reservoir thins out slightly and opens into a space between the wall of the ejaculatory duct and the aedeagus, right next to the basiphallus (Fig. 6D). This gland has a very similar shape to that of all other studied tephritid species.

The length of the aedeagus is variable among the species of *Anastrepha* and is probably correlated with the length of the female terminalia in this genus (Norrbom, personal communication). Although a statistical analysis was not performed, for some species aedeagus length was measured and compared with average aculeus length. We found that the species with a long aedeagus has a long aculeus as well. For instance, in *A. fraterculus* and *A. obliqua,* the aculeus is 1.4–1.8 mm long, while the aedeagus is 2.38–2.72 mm long. In contrast, species with long aculeus such as *A. cordata* (4.7–5.5 mm) and *A. ludens* (3.2–5 mm) have an aedeagus length of approximately 6.18 and 5.71 mm, respectively.

The male terminalia involves structures such as the epandrium, proctiger and the inner and outer surstyli. The epandrium is a rigid and very sclerotized structure of semi-spherical shape. In its medial posterior region lies the membranous proctiger, which can be retracted or expanded. The surstyli rise out of the inferior base of the epandrium; both the inner and outer surstyli are partially fused together.

The most important differences among males were observed in structures such as the accessory glands, ejaculatory apodeme, distiphallus and male terminalia.

In the species examined, the accessory glands varied in number, shape and size. *Anastrepha serpentina* has six pairs, *A. striata* four pairs and *A.cordata* only two pairs. In the species of the *fraterculus* group (cf. *A. ludens, A. obliqua* and *A. fraterculus*), seven pairs were observed. Forked long glands are found in *A. serpentina, A.cordata, A. ludens, A. obliqua* and *A. fraterculus*; only *A. striata.* has one pair of simple long glands. Forked medium glands were observed only in *A. serpentina, A. striata, A. obliqua* and *A. fraterculus.* All species, however, have simple medium

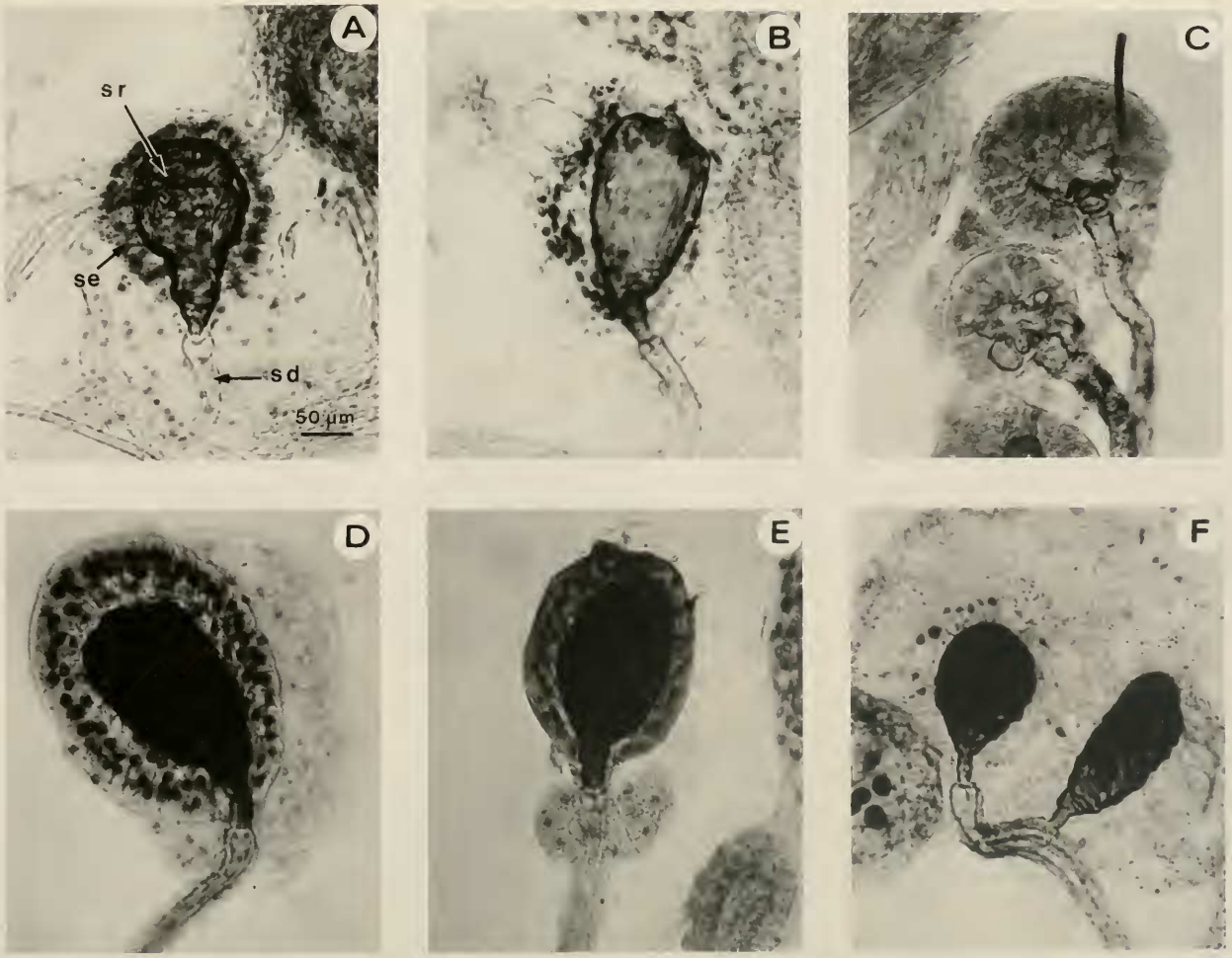

Fig. 3. Microphotographs of the spermathecae (Feulgen-light green). A, *Anastrepha serpentina*. B, *A. striata*. C, *A. cordata*. D, *A. ludens*. E, *A. obliqua*. F, *A. fraterculus*. Abbreviations: se = spermathecal epithelium; spd = spermathecal duct; sc = spermathecal capsule.

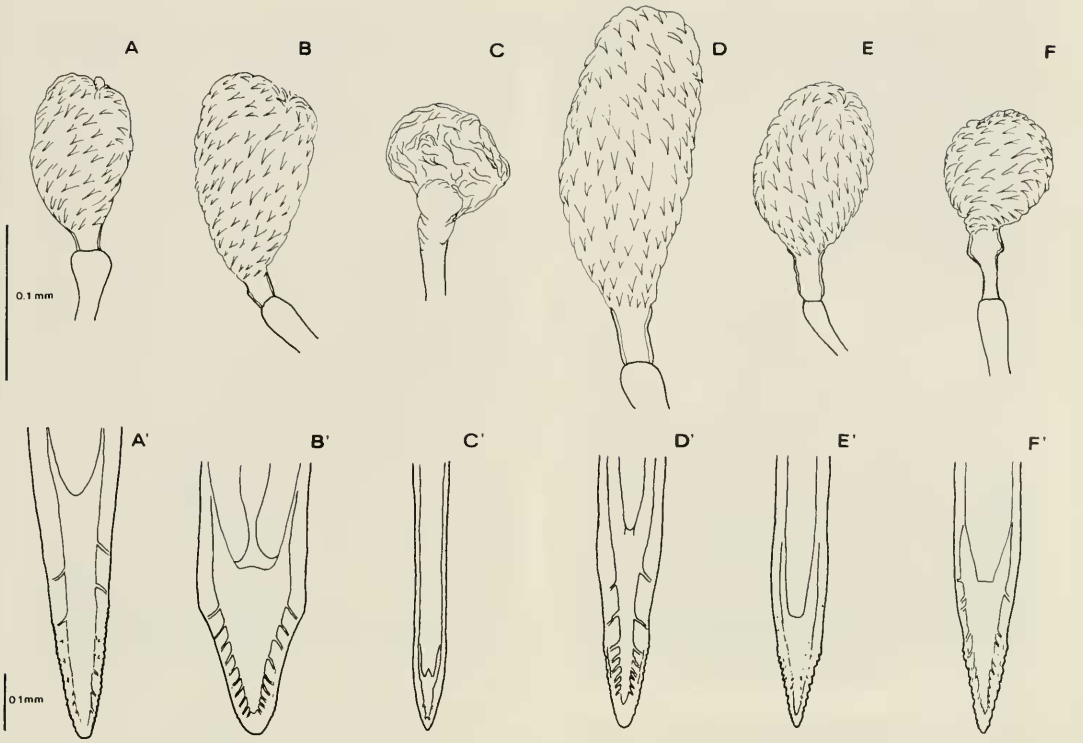

Fig. 4. Outlines of the spermathecal capsules and aculeus tips. A,A', *Anastrepha serpentina*. B, B', *A. striata*. C, C', *A. cordata*. D, D', *A. ludens*. E, E', *A. obliqua*. F, F', *A. fraterculus*.

glands, although in varying number: there is one pair in *A. cordata, A. obliqua,* and *A. fraterculus,* two pairs in *A. striata* and *A.ludens* and four pairs in *A. serpentina.* Only in *A. ludens, A. obliqua* and *A. fraterculus* are there four pairs of small glands. The same number, shape and size of glands are found only in *A. obliqua* and *A. fraterculus.* (Fig. 7, Table 4).

In the mature males studied, the ejaculatory apodeme tends to be narrow in *A. ludens, A. fraterculus,* and *A. striata,* whereas in the others, especially *A. cordata,* is visibly wider at its apical half (Fig. 8). However, the differences should be considered with some reserve, due to the fact that intraspecific size can vary depending on the degree of maturity of each individual (Drew 1969). To study this properly, careful comparison must be made of each species over time, and the results presented should include a range of intraspecific variation.

In general, the distiphallus (Fig. 9) has a bulky basal portion known as the basal lobe, with tiny microspines on its surface. The rest is a membranous, semitransparent unit with an interior sclerotized duct. All *Anastrepha* and *Toxotrypana* species present an internal apical sclerite "T" shaped (sensu Norrbom 1985).

Within the male terminalia, some interspecific differences stand out, particularly the shape of the outer surstyli when viewed laterally. In most of the studied species, they were elongated, except in *A. cordata* in which the surstyli are very short and widened at the base (Fig. 10).

Discussion

The general anatomy of the female reproductive system is similar in all of the studied *Anastrepha* species, as well as other Tephritidae such as *Rhagoletis pomonella* (Dean 1935), *C. capitata* (Hanna 1938), *B.*

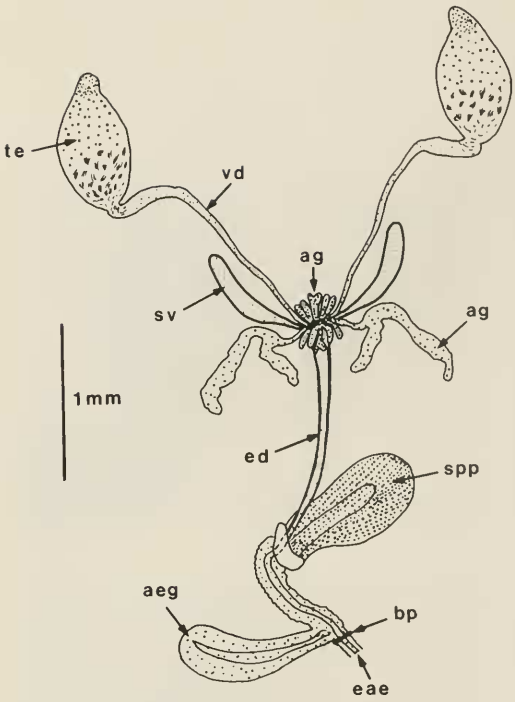

Fig. 5. General morphology of the reproductive system in the male of *Anastrepha serpentina.* Abbreviations: aeg = aedeagal gland; ag = accessory gland; bp = basiphallus; eae = entrance of the ejaculatory duct to aedeagus; ed = ejaculatory duct; spp = sperm pump; sv = seminal vesicles; te = testis; vd = vas deferens.

tryoni (Drew 1969) and *B. oleae* Gmelin (Solinas and Nuzzaci 1984).

The number of ovarioles present in the six species of *Anastrepha* varies between 20 and 33 per ovary. This number is also very similar to that of other species of fruit flies such as *C. capitata,* which has an average of 28 ovarioles (Hanna 1938), *R. pomonella* with an average of 24 (Dean 1935), and *Bactrocera tryoni* with about 22 (Anderson and Lyford 1965). However, species such as *R. conversa* and *R. nova* have an average of 9 and 12 ovarioles, respectively (Flores et al. 1987).

In all *Anastrepha* females, the ovarian follicle contains the oocyte and 15 nutritive cells, similar results were reported previously for *A. obliqua* (Bressan and Costa Teles 1991) and other species such as *B. tryoni* (Anderson and Lyford 1965), *R. conversa* and *R. nova* (Flores et al. 1987).

The anatomy of the vagina in the Tephritidae, except in the case of *R. pomonella* (Dean 1935), has never been clearly described. In *C. capitata* the vagina and vaginal duct were depicted without any mention of the *bursa copulatrix* and the ventral receptacle (Hanna 1938, Guillén 1983). The ventral receptacle was illustrated as an unpaired, sclerotized organ in *X. unipuncta* by Souza Lopes (1939) as well as in other groups of Tephritidae (Munro 1984). In *B. tryoni* this structure was termed the morula gland by Drew (1969).

In the few species of *Anastrepha* studied previously, neither the *bursa copulatrix* nor the ventral receptacle has been adequately described. With respect to *A. suspensa* and *A. ludens,* the posterior region of the vagina was mislabeled the oviduct by Dodson (1978), while Servín-Villegas and Jiménez-Jiménez (1995) believed that the vagina is formed by a widening in the oviduct. In *A.*

Table 4. Comparison of some characteristics of the accessory glands of males of six *Anastrepha* species.

Species	Accessory glands (pairs)					
	Long		Medium			
	Forked	Simple	Forked	Simple	Small	Total
A. serpentina	1	—	1	4	—	6
A. striata	—	1	1	2	—	4
A. cordata	1	—	—	1	—	2
A. ludens	1	—	—	2	4	7
A. obliqua	1	—	1	1	4	7
A. fraterculus	1	—	1	1	4	7

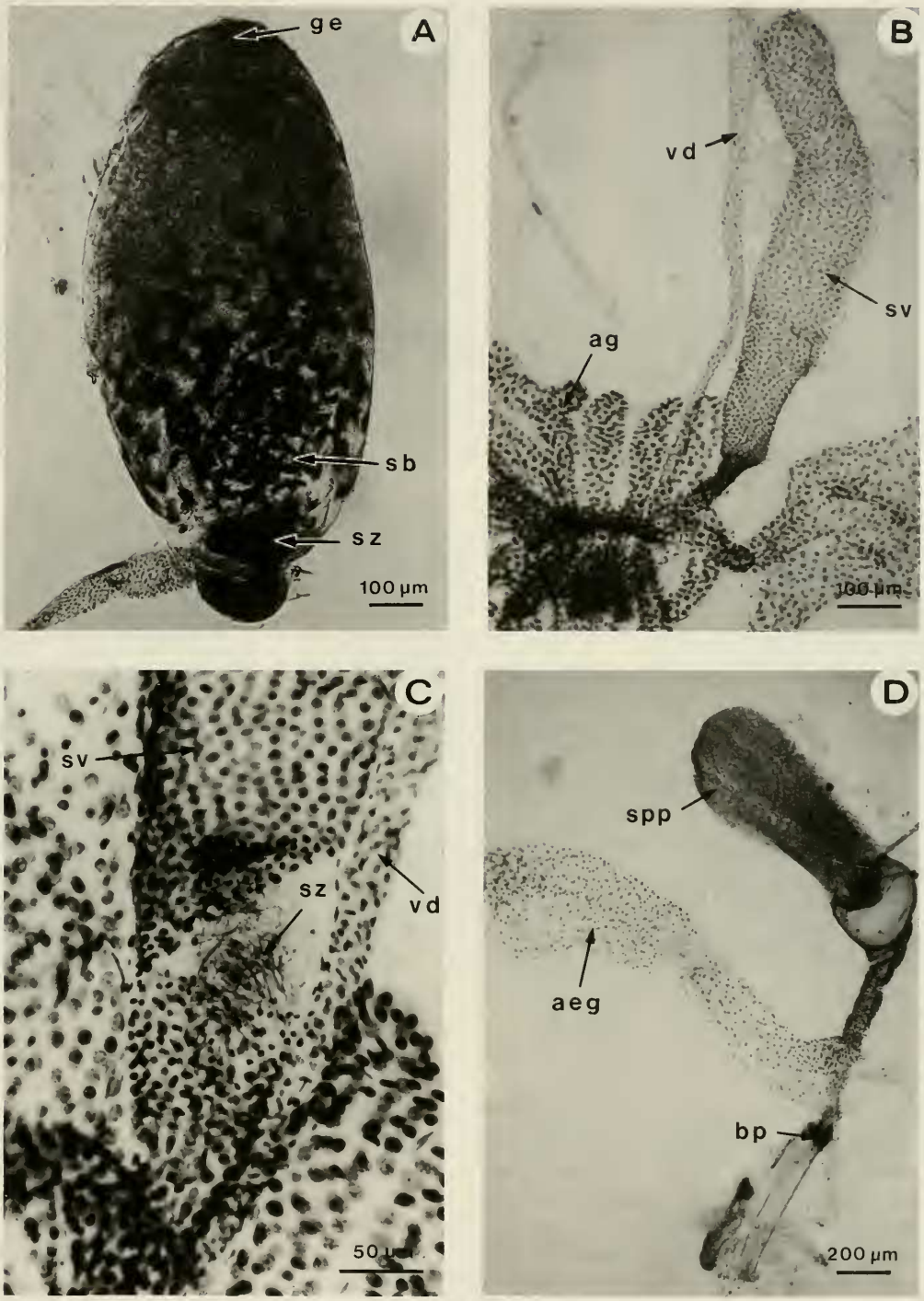

Fig. 6. Microphotographs of some structures of the reproductive system in the male of *Anastrepha serpentina* (Feulgen-light green). A, Testis. B, Accessory glands. C, Seminal vesicles and vas deferens. D, Sperm pump and aedeagal gland. Abbreviations: aeg = aedeagal gland; ag = accessory glands; bp = basiphallus; sb = sperm bundles; spp = sperm pump; sv = seminal vesicles; sz = spermatozoids; vd = vas deferens.

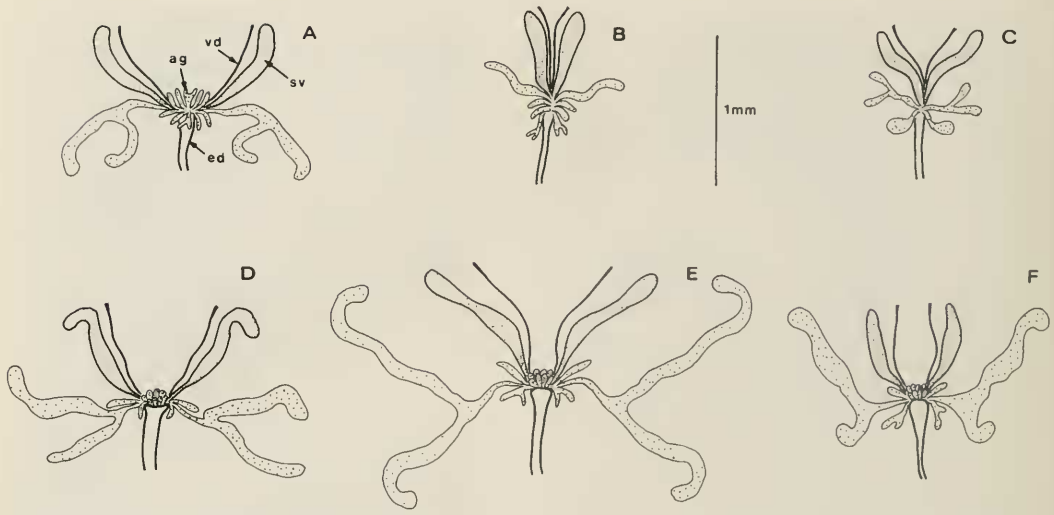

Fig. 7. Outlines of the accessory glands and seminal vesicles. A, *Anastrepha serpentina.* B, *A. striata.* C, *A. cordata.* D, *A. ludens.* E, *A. obliqua.* F, *A. fraterculus.* Abbreviations: ag = accessory glands; ed = ejaculatory duct; sv = seminal vesicles; vd = vas deferens.

serpentina, only the anterior vagina and vaginal duct have been recognized (Martínez et al. 1995).

Other authors such as Dean (1935) and Williamson (1989) characterize the vagina as consisting of two regions: the anterior, which is made up of the *bursa copulatrix* as well as the ventral receptacle, also called the fertilization chamber by Solinas and Nuzzaci (1984). The second is the posterior region, which consists of the vaginal duct.

In insects the *bursa copulatrix* acts as a receptacle for the male intromittent organ (Snodgrass 1935). In *Anastrepha* it receives the distiphallus during copula, but spermatozoids are deposited in the ventral receptacle before being transferred to the spermathecae. The few which remain in the ventral receptacle will be the first spermatozoids to fertilize the eggs (Dean 1935, Solinas and Nuzzaci 1984).

The presence of three spermathecae in all species of *Anastrepha* is characteristic of most Trypetinae (*sensu* Hancock 1986a), except in some species of *Rhagoletis* such as *R, nova* and *R. conversa* (Flores et al. 1987), *R.striatella* Wulp (Bush 1966) and other Carpomyina (Norrbom 1994). In contrast, Dacinae species such as *C. capitata, B. tryoni* and *B. oleae* have only two spermathecae (Hanna 1938, Drew 1969, Solinas and Nuzzaci 1984), as well as all Tephritinae (Hancock 1986b).

Female accessory glands in tephritid females have been misrepresented as colleterial glands by some authors (Hanna 1938, Drew 1968, Flores et al. 1987). This term is employed specifically for the accessory glands of the Orthoptera, in which the colleterial glands are responsible for secreting substances which form the ootheca (Davey 1985a). In Diptera, the function of the accessory glands is poorly understood, except in *Glossina* (Glossinidae), in which they are called milk glands because their secretions serve as nourishment for the intrauterine larvae (Matzuda 1976, Davey 1985a).

For tephritids, the function of the accessory glands is not well understood, although Christenson and Foote (1960) speculated that their secretions act as a vaginal lubricant before oviposition. Solinas and Nuzzaci (1984) mentioned that these secretions can help carry sperm toward the fertilization chamber. It has also been suggested that the accessory glands produce

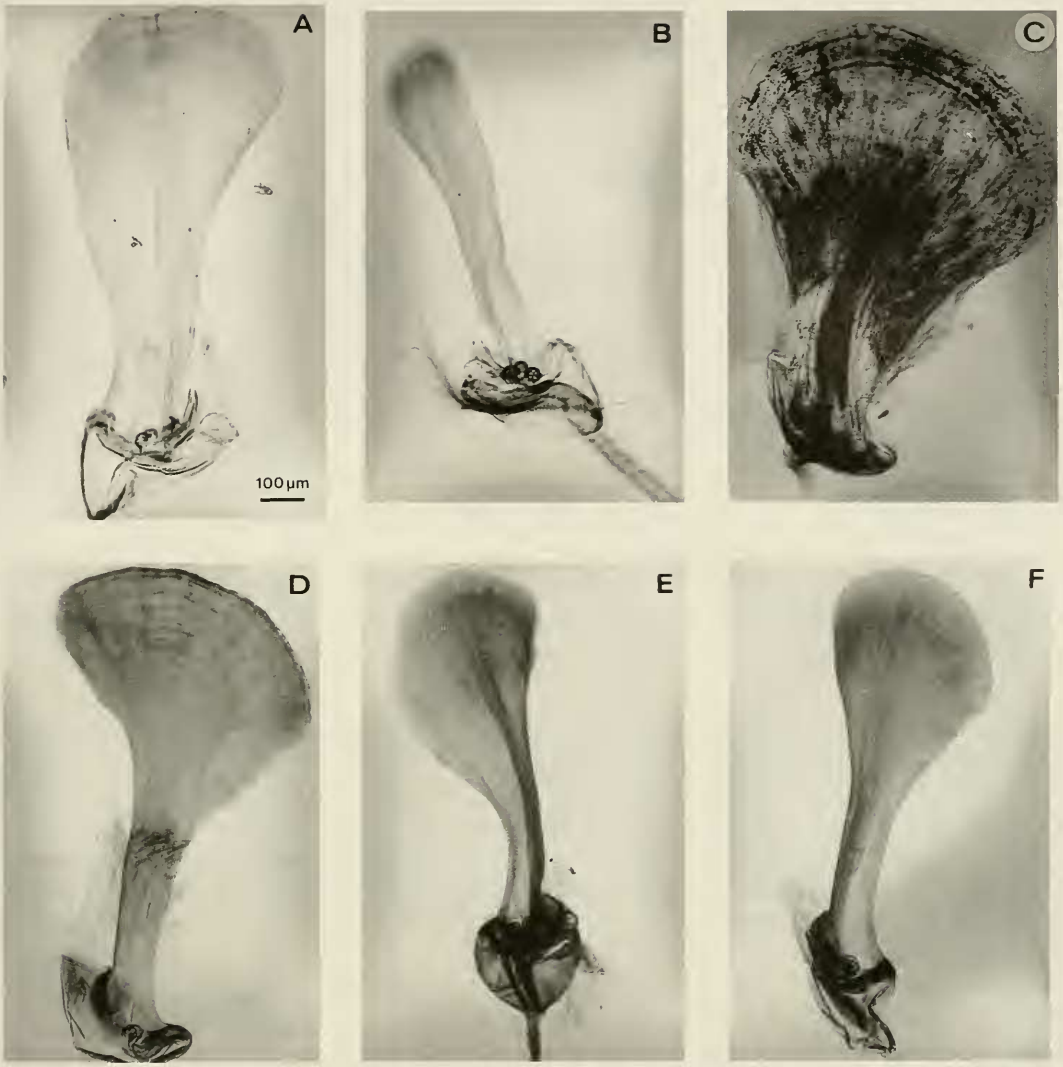

Fig. 8. Microphotographs of ejaculatory apodemes of the sperm pump (chlorazol black E). A, *Anastrepha serpentina*. B, *A. striata*. C, *A. cordata*. D, *A. ludens*. E, *A. obliqua*. F, *A. fraterculus*.

marking pheromones deposited by the female after oviposition so that other females recognize infested fruit (Prokopy and Roitberg 1984).

The general anatomy of the male reproductive system for *Anastrepha* is similar in all studied species as well as other Tephritidae (Hanna 1938, Souza Lopes 1939, Drew 1969).

As to the location of the seminal vesicles in Tephritidae, various authors (Anwar 1971, Williamson 1989, Bressan 1995, Servín-Villegas and Jiménez-Jiménez 1995) stated that they are located to the basal part of the testis perhaps due to the abundant presence of free spermatozoids. In *C. capitata* they confused the seminal vesicles with the anterior part of the ejaculatory duct (Hanna 1938). As to *X. unipunctata,* Souza Lopes (1939) stated that the seminal vesicles are two structures which open into the anterior region of the ejaculatory duct near the vas deferens.

In mature males of *Anastrepha,* free sper-

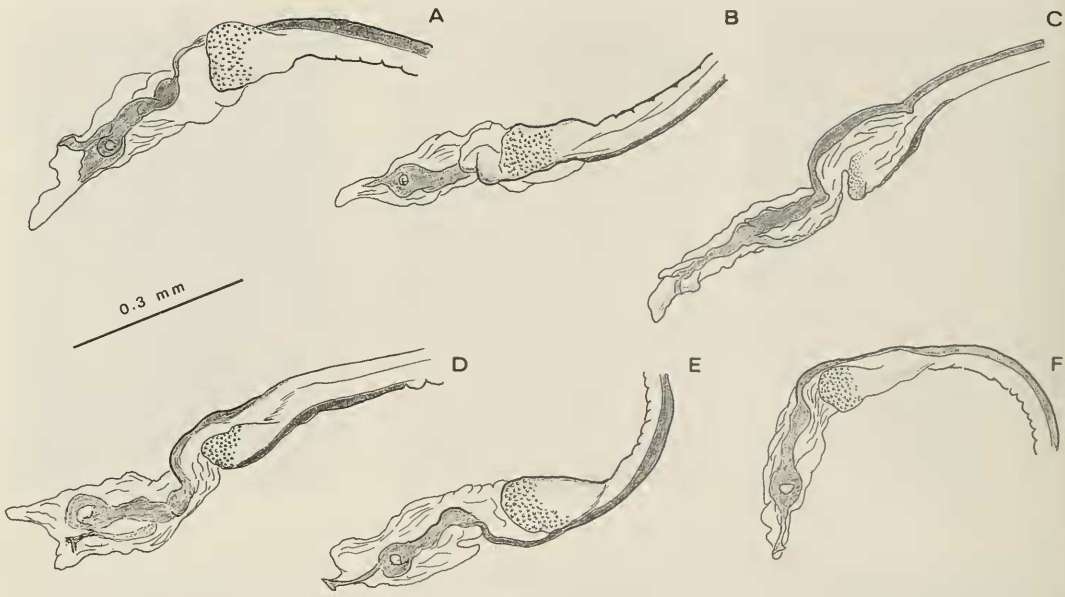

Fig. 9. Outlines of distiphallus. A, *Anastrepha serpentina.* B, *A. striata.* C, *A. cordata.* D, *A. ludens.* E, *A. obliqua.* F, *A. fraterculus.*

matozoids were observed at the base of the testes, all along the vas deferens and in the seminal vesicles. In this genus the seminal vesicles open into the ejaculatory duct parallel to the vas deferens and between the accessory glands, as is the case in other groups of Diptera (see Matzuda 1976). In *Anastrepha* the seminal vesicles have been confused with the accessory glands due to their proximity to them and because no spermatozoids have been observed in the seminal vesicles.Their function for the storage of the mature spermatozoids until the moment of copula is well known in different species of insects (Snodgrass 1935, Davey 1985b).

The accessory glands of *Anastrepha* males has not been described in detail previously. It is quite probable that they are responsible for secreting substances which form the seminal liquid which accompany the spermatozoids; this has been documented for other groups of insects (Davey 1985b). The species examined from the *fraterculus* group all had seven pairs of accessory glands, the highest number observed. The other species had from two to six pairs, and the number differing from species to species (Table 3). In *A. suspensa,* which also belongs to the *fraterculus* group, the existence of three to four pairs was mentioned by Dodson (1978), It is probable that this author did not observe the four pairs of small glands because they are difficult to discern without dye; he may have mistaken one pair for seminal vesicles. For *A. ludens,* Servín-Villegas and Jiménez-Jiménez (1995) reported the presence of four pairs, which in all likelihood correspond to the pair of long glands, two pairs of medium glands and one pair of seminal vesicles. The four small glands were not observed (Table 4).

Xanthaciura unipuncta has only two pairs of accessory glands (Souza Lopes 1939). For *C. capitata,* Hanna (1938) and Valdés Carrasco and Prado Beltrán (1990) described the presence of four pairs, one very long and three short, although we assume that one pair is actually the seminal vesicles. For *B. tryoni,* Drew (1969) described 4 pairs, but one of them also exhibits marked morphological differences which

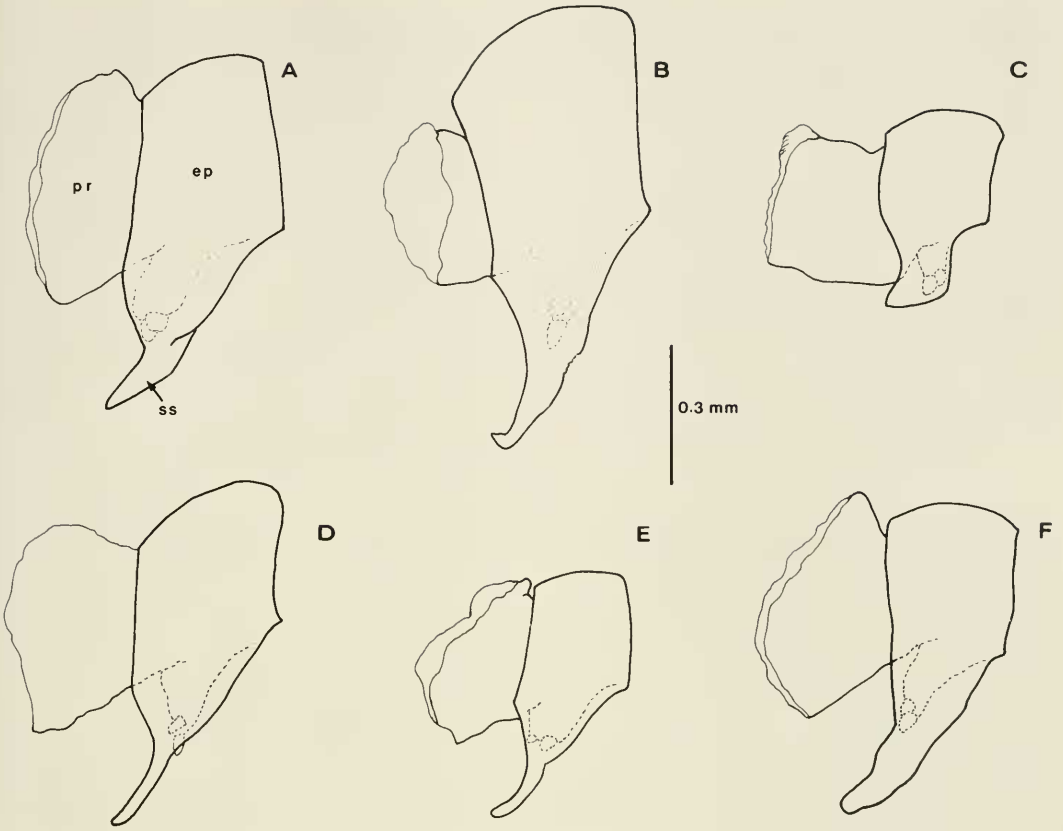

Fig. 10. Simplified outlines of the male genitalia in lateral view. A, *Anastrepha serpentina*. B, *A. striata*. C, *A. cordata*. D, *A. ludens*. E, *A. obliqua*. F, *A. fraterculus*. Abbreviations: ep = epandrium; pr = proctiger; ss = outer surstyli.

make us believe that they are seminal vesicles.

The sperm pump is made up of a semispherical base to which a sclerotized internal structure called the ejaculatory apodeme is attached; many muscles are inserted onto the apodeme. The sperm pump pumps seminal liquid and distends the aedeagus during copula (Matzuda 1976).

In this respect, most studies of Tephritidae call the sperm pump the ejaculatory apodeme, while other authors such as Hanna (1938) and Drew (1969) call it the erecting and pumping organ.

In all *Anastrepha* species examined, the aedeagal gland corresponds to the oval-shaped gland described for *C. capitata* by Hanna (1938), and to the second accessory gland indicated by Bressan (1995) for *Anastrepha* spp. Given its glandular anatomical characteristics and because it opens into a region between the ejaculatory duct and the aedeagus at the base of the basiphallus, its secretions must empty into this space. The secretions may function as a lubricant, as suggested by Hanna (1938), and may also serve to distend the aedeagus during copula.

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Literature Cited

Anderson, D. T. and G. C. Lyford. 1965. Oogenesis in *Dacus tryoni* (Frogg) (Diptera: Trypetidae). Australian Journal of Zoology 13:423–435.

Anwar, M., D. L. Chambers, K. Ohinata and R. M. Kobayashi. 1971. Radiation sterilization of the Mediterranean fruit fly (Diptera: Tephritidae): Comparison of spermatogenesis in flies treated as pupae or adults. Annals of the Entomological Society of America 64:627–633.

Bressan, S. 1995. Desenvolvimento e potencial reprodutivo do macho de *Anastrepha spp.* (Diptera: Tephritidae) em condicoes naturais. Revista Brasileira de Entomologia 39(4):849–854.

Bressan, S. and M. Costa Teles. 1991. Ovogènese em *Anastrepha obliqua* (Macquart) (Diptera, Tephritidae). Revista Brasileira de Entomologia 35(4): 691–696.

Bush, G. L. 1966. The taxonomy, cytology and evolution of the genus *Rhagoletis* in North America (Diptera, Tephritidae). Bulletin of the Museum of Comparative Zoology 134(11):431–560.

Carayon, J. 1969. Emploi du noir chlorazol en anatomie microscopique des insectes. Annales de la Société Entomologique de France (N.S.) 5(1): 179–193.

Christenson, L. D. and R. H Foote. 1960. Biology of fruit flies. Annual Review of Entomology 5:171–192.

Dampf, A. 1933. Estudio sobre el oviscapto de las moscas de la fruta (*Anastrepha spp.*) de México. Irrigación en México (Comisión Nacional de Irrigación de la Secretaría de Agricultura y Fomento) 6(3):253–265.

Davey, K. G. 1985a. The female reproductive tract, pp. 15–36. *In* Kerkut G. A. and L.I. Gilbert, eds. Comprehensive Insect physiology, biochemistry and pharmacology. Pergamon Press. Vol.I.

Davey, K. G. 1985b. The male reproductive tract, pp. 1–14. *In* Kerkut G. A. and L.I. Gilbert, eds. Comprehensive Insect physiology, biochemistry and pharmacology. Pergamon Press. Vol.I.

Dean, R. W. 1935. Anatomy and postpupal development of the female reproductive system in the apple maggot fly, *Rhagoletis pomonella* Walsh. New York State Agricultural Experimental Station Technical Bulletin 229:1–31.

Dodson, G. 1978. Morphology of the reproductive system in *Anastrepha suspensa* (Loew) and notes on related species. The Florida Entomologist 61(4):231–239.

Drew, R. A. I. 1969. Morphology of the reproductive system of *Strumeta tryoni* (Froggatt) (Diptera: Trypetidae) with a method of distinguishing sexually mature adult males. Journal of the Australian Entomological Society 8:21–32.

Flores, R. V., D. Frías and A. H. Martínez. 1987. Histología y morfología del sistema reproductor en hembras de *Rhagoletis conversa* (Brethes) y de *Rhagoletis nova* (Schiner) (Diptera, Tephritidae). Revista Brasileira de Zoologia 4(1):33–46.

Gabe, M. 1968. Techniques histologuiques. Ed. Masson et Cie. 1113 pp.

Guillén, A. J. C. 1983. Manual para la diferenciación de moscas del mediterraneo *Ceratitis capitata* (Wied.) silvestres (fértiles) de moscas irradiadas (estériles). Programa Mosca del Mediterráneo, SARH. 102 pp.

Hancock, D. 1986a. Classification of the Trypetinae (Diptera: Tephritidae) with a discussion of the Afrotropical fauna. Journal of Entomological Society of South Africa 49:275–305.

Hancock, D. 1986b. New genera and species of African Tephritinae (Diptera≡phritiade), with comments on some currently unplaced of misplaced taxa and on classification. Transactions of the Zimbabwe Scientific Association 63:16–34.

Hanna, A. D. 1938. Studies on the Mediterranean fruit fly: *Ceratitis capitata* (Wied.) I. The structure and operation of the reproductive organs. Bulletin de la Societé Foread ler De Entomologie 22:39–52.

Hernández-Ortiz, V. and M. Aluja. 1993. Listado de especies del género neotropical *Anastrepha* (Diptera: Tephritidae) con notas sobre su distribución y plantas hospederas. Folia Entomológica Mexicana 88:89–105.

Martínez, M. I., V. Hernández-Ortiz and L. R. Luna. 1995. Desarrollo sexual en *Anastrepha serpentina* (Wiedemann) (Diptera: Tephritidae). Acta Zoológica Mexicana (n.s.) 65:75–88.

Matzuda, R. 1976. Morphology and evolution of the insect abdomen. Pergamon, Press. pp. 335–363.

McAlpine J. F. 1981. Morphology and terminology—adults, pp. 9–63. *In* McAlpine J. F., B. V. Peterson, G. E. Shewell, H. J. Teskey, J. R. Vockeroth and D. M. Wood, eds. Manual of Nearctic Diptera. Research Branch Agriculture Canada. Monograph 27.

Munro, H. K. 1984. A taxonomic treatise on the Dacidae (Tephritoidea, Diptera) of Africa. Entomology Memoirs. Department of Agriculture and Water Supply. Republic of South Africa. 61, ix + 1–313.

Norrbom, A. L. 1985. Phylogenetic analysis and taxonomy of the *cryptostrepha, daciformis, robusta* and *schausi* species groups of *Anastrepha* Schiner (Diptera: Tephritidae). Pennsylvania State University (Ph.D. Thesis). 355 pp.

Norrbom, A. L. and K. C. Kim. 1988. Revision of the *schausi* Group of *Anastrepha* Schiner (Diptera, Tephritidae), with a Discussion of the Terminology of the female terminalia in the Tephritoidea. Annals of the Entomological Society of America 81(2):164–173.

Norrbom, A. L. 1994. New species and phylogenetic analysis of *Cryptodacus, Haywardina,* and *Rhagoletotrypeta* (Diptera: Tephritidae). Insecta Mundi 8(1–2):37–65.

Prokopy, R. J. and B. D. Roitberg. 1984. Foraging behavior of true fruit flies. American Scientist 72: 41–49.

Servín-Villegas, R. and M. L. Jiménez-Jiménez. 1995. El aparato reproductor y ciclo gonádico de la mosca de la fruta *Anastrepha ludens.* Southwestern Entomologist 20(2):223–227.

Snodgrass, R. E. 1935. Principles of insect morphology. McGraw-Hill Book Company. 667 pp.

Solinas, M. and G. Nuzzaci. 1984. Functional anatomy of *Dacus oleae* Gmel. female genitalia in relation to insemination and fertlization processes. Entomologica, XIX Bari 16(12):135–165.

Souza Lopes, H. de. 1939. Sobre a *Xanthaciura unipuncta* Malloch, 1933 (Dipt.: Trypetidae), pp. 424–431. *In* Livro de Homenagem aos Prof. Alvaro e Miguel Ozorio de Almeida, Rio de Janeiro, Brasil.

Valdés-Carrasco, J. and E. Prado-Beltrán. 1990. Esqueleto y musculatura de la mosca del mediterraneo, *Ceratitis capitata* (Wiedemann) (Diptera: Tephritidae). Folia Entomologica Mexicana 80: 59–225.

Williamson, D. L. 1989. Oogenesis and spermatogenesis, pp. 141–151. *In* Robinson A. S. and Hooper, G., eds., Fruit flies. Their biology, natural enemies and control Vol. 3A.